

21. (Amended) Process according to claim 16, wherein said hypotonic treatment in step iii) is obtained by means of a hypotonic solution comprising:

NaCl in concentrations lower than 0.12 M; and optionally sodium citrate in concentrations lower than 0.025 M.

REMARKS

It is respectfully requested that the examination of this application proceed on the basis of the amendatory action taken herein and that this amendment be entered prior to calculating the filing fee and according the application a filing date.

Attached hereto is a Marked-Up Version of the Changes Made to the claims by the current Pre-Examination Amendment. The attached page is captioned "Marked-Up Version to Show Changes Made".

The issuance of a Notice of Allowance is respectfully solicited.

Please charge any fees which may be due and which have not been submitted herewith to our Deposit Account No. 01-0035.

Respectfully submitted,

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Docket No.: 205,360

MARKED-UP VERSION TO SHOW CHANGES MADE BY AMENDMENT

Claims 8, 9, 13-16, and 18-21 have been amended as follows:

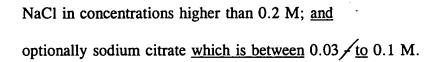
- 8. (Amended) Inactivated microorganisms according to claims 1—7, wherein said microorganism is Saccharomyces cerevisiae.
- 9. (Amended) Food compositions characterized in that wherein the inactivated microorganisms according to claims 1 -8 are used.
- 13. (Amended) Use of one or more inactivated micoorganisms according to claims 1 -12 in human or animal alimentation.
- 14. (Amended) Use of the microorganisms according to claims 1 -12 as components of feeding or premixes in zootechnics.
- 15. (Amended) Use of the microorganisms according to claims 10 -12 for the feeding of fish.
- 16. (Amended) Process for the preparation of inactivated microorganisms containing one or more soluble and/or solubilizable substances having pharmacological activity and/or nutritional substances having pharmacological activity, according to claims-1 -12 comprising the following steps:
- (i) drawing out the endocellular mass of a suitable microorganism by means of hypertonic treatment, separation of the drawn out endocellular mass and recovery of the empty microorganisms;
 - (ii) optional inactivation of the microorganism obtained in

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Step i) chemically or physically, leaving the external membrane of the microorganism unaltered; and

- (iii) intracellular loading of one or more soluble and/or solubilizable substances having pharmacological activity and/or nutritional substances having pharmacological activity, into the inactivated microorganism obtained in Step i) or Step ii), is obtained by means of hypo- and/or iso-tonic treatment.
- 18. (Amended) Process according to claims 16 and 17 characterized in that wherein step iii) is followed by treatment of the microorganisms with a fixative or a disinfectant agent.
- 19. (Amended) Process for the preparation of inactivated microorganisms described in according to claims 1 -12 which comprises the following steps:
- 1) <u>inactivating</u> the microorganism is inactivated by thermal treatment, at 60-65°C for 30-120 min;
- 2) <u>resuspending</u> the inactivated cells of the microorganism are resuspended in an isotonic medium comprising the active principle to be incorporated;
 - 3) <u>stirring</u> the suspension is left-stirring for 48-72 hours;
 - 4) <u>centrifuging</u> the suspension is centrifuged;
- 5) optional buffering and/or fixation is carried out using formalin and/or glutaraldehyde.
- 20. (Amended) Process according to claims-16 and 17 characterized in that wherein the hypertonic treatment in step i) is obtained by incubation of a hypertonic solution comprising:



21. (Amended) Process according to claims 16 and 17, in which wherein said hypotonic treatment in step iii) is obtained by means of a hypotonic solution comprising:

NaCl in concentrations lower than 0.12 M; and

optionally sodium citrate in concentrations lower than 0.025 M.